Reference 1

Invitrogen life technologies

SuperScript™ II Reverse Transcriptase

Reliable RT for Full-length cDNA Synthesis and High Yields

Description:
SuperScript™ II Reverse Transcriptase (RT) is an improved version of SuperScript™ RT. It is a DNA polymerase that synthesizes a complementary DNA strand from single-stranded RNA, DNA, or an RNA:DNA hybrid. This enzyme is genetically engineered by the introduction of point mutations rather than a deletion in the RNase H active center. Like SuperScript™ RT, SuperScript™ II RT has reduced RNase H activity. Unlike SuperScript™ RT, however, the selective mutations within the RNase H domain maintain full polymerase activity. This structural modification eliminates degradation of RNA molecules during first-strand cDNA synthesis and gives SuperScript™ II RT superior performance characteristics, including:

- Greater first-strand cDNA yields
- More full-length cDNA synthesis
- Full activity at 42°C

Source: Purified from *E. coli* expressing the *pol* gene of M-MLV (1,2), mutagenized to reduce the RNase H activity.

Performance and Quality Testing: SDS-PAGE purity; endodeoxyribonuclease, exodeoxyribonuclease, and ribonuclease assays; and yield and length of cDNA product.

Unit Definition: One unit of SuperScript™ II is the amount of enzyme required to incorporate 1 nmole of eoxyribonucleotide into acid-precipitable material in 10 min. at 37°C using poly(A)•oligo(dT)12-18 as template•primer (3).

Product	Cat. No.	Size	Conc.	Price	Select
SuperScript™ II Reverse Transcriptase	18064-022	2,000 units	200 units/µl	60.00	☐
SuperScript™ II Reverse Transcriptase	18064-014	10,000 units	200 units/µl	220.00	☐
SuperScript™ II Reverse Transcriptase	18064-071	4 x 10,000 units	200 units/µl	792.00	☐

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Related Products

10 mM dNTP Mix and Individual 10 mM dNTPs
100 mM dNTP Set and Individual 100 mM dNTPs
2.5 mM dNTP Mix
Platinum Pfx DNA Polymerase
Platinum Taq DNA Polymerase
SuperScript Plasmid System for cDNA Synthesis and Cloning
Taq DNA Polymerase

Technical Information

Description	Document Type
SuperScript II RT Brochure	Brochure
SuperScript II Reverse Transcriptase	Manual

Unit Reaction Conditions: 50 mM Tris-HC1 (pH 8.3), 40 mM KC1, 6 mM MgC1 2 , 1 mM DTT, 0.5 mM [^{3}H]dTTP, 0.1 mM poly(A), 0.1 mM oligo(dT)$_{12-18}$, 0.1 mg/m1 BSA, and enzyme in 50 µl for 10 min. at 37°C.

Contents and Storage:
SuperScript ← II is supplied with a vial of 5X first-strand buffer [250 mM Tris-HCl (pH 8.3), 375 mM KCl, 15 mM $MgCl_2$], and a vial of 100 mM DTT. Store at -20°C. Guaranteed stable for 6 months when properly stored.

Reference(s):
1. Kotewicz, M. *et al.* (1992) *Gene* **35:** 249.
2. Gerard, G.F. *et al.* (1986) *DNA* **5:** 271.
3. Houts, G. *et al.* (1979) *J. Virol* **29:** 519.

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SuperScript™ II Reverse Transcriptase

Cat. Nos.	Size:
18064-022	2,000 units
18064-014	10,000 units
18064-071	4 x 10,000 units
Conc: 200 U/µl	Store at -20°C (non-frost-free)

Description

SuperScript™ II Reverse Transcriptase is an engineered version of M-MLV RT with reduced RNase H activity and increased thermal stability. The enzyme is purified to near homogeneity from *E. coli* containing the modified *pol* gene of Moloney Murine Leukemia Virus (1,2). The enzyme can be used to synthesize first-strand cDNA at higher temperatures than conventional M-MLV RT, providing increased specificity, higher yields of cDNA, and more full-length product. It can generate cDNA up to 12.3 kb.

Components

SuperScript™ II RT, 5X First-Strand Buffer (250 mM Tris-HCl, pH 8.3 at room temperature; 375 mM KCl; 15 mM $MgCl_2$), 0.1 M DTT

Storage Buffer

20 mM Tris-HCl (pH 7.5), 100 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 0.01% (v/v) NP-40, 50% (v/v) glycerol

Storage Conditions

Store all components at -20°C in a non-frost-free freezer.

Thaw 5X First-Strand Buffer and 0.1 M DTT at room temperature just prior to use and refreeze immediately.

Unit Definition

One unit incorporates 1 nmole of dTTP into acid-precipitable material in 10 min. at 37°C using poly(A)•oligo$(dT)_{25}$ as template-primer (3).

For general laboratory use. FOR *IN VITRO* USE ONLY.

Transcriptor Reverse Transcriptase

Cat. No. 03 531 317 250 U for 25 reactions
Cat. No. 03 531 295 500 U for 50 reactions
Cat. No. 03 531 287 2 000 U (4 x 500 U) for 200 reactions

Version 1, June 2003
Store at −15 to −25°C

Product overview

Pack content

Vial	Label	Content
RT red cap	Transcriptor Reverse Transcriptase (20 U/µl)	• 12,5 µl (250 U pack size) • 25 µl (500 U pack size) • 4 x 25 µl (2 000 U pack size) Storage buffer: 200 mM potassium phosphate, 2 mM dithiothreitol, 0.2% Triton X-100 (v/v), 50% glycerol (v/v), pH approx. 7.2
RT buffer 5x colorless cap	Transcriptor RT Reaction Buffer (5x)	• 1 ml (250 U pack size and 500 U pack size) • 2 x 1 ml (2 000 U pack size) 5 x conc.: 250 mM Tris/HCl, 150 mM KCl, 40 mM $MgCl_2$, pH approx. 8,5 (25°C)

Product description

Transcriptor Reverse Transcriptase is a fast new recombinant reverse transcriptase expressed in *E. coli*. It includes a RNA-directed DNA polymerase, a DNA-dependent DNA polymerase, a RNase H and an unwinding activity. As template ss DNA and ss RNA are accepted in the presence of a primer.

Transcriptor Reverse Transcriptase transcribes long RNAs and is recommended for RT-PCR because of its high sensitivity in connection with high thermostability. Transcriptor Reverse Transcriptase is also recommended for GC-rich templates and for labeling reactions during cDNA synthesis.

Enzyme properties

Volume activity	20 U/µl
Specific activity	50 U/µg
Source	New recombinant reverse transcriptase expressed in *E. coli*
Divalent ion requirement	Mg^{2+}
Product size of reverse transcription reaction	Up to 14 kb
RNase H activity	yes
Incorporation of modified nucleotides	Accepts labeled nucleotides like DIG-, Biotin-, Cy3-, Cy5- and aminoallyl-dUTP. For reverse transcription reaction in array applications (*e.g.* aminoallyl technique) a reaction temperature of 42°C is recommended and results in lower gene to gene variations than at 55°C.
Template	Accepts total RNA and mRNA from tissues, cell lines or blood, viral RNA, *in vitro* transcribed RNA
Prevention of carry-over contamination	Compatible with PCR reactions where dUTP is incorporated
Purity (SDS-PAGE)	≥ 90%
Bioburden	≤ 50 cfu/ml
Inactivation	After incubation for 5 min at 85°C Transcriptor Reverse Transcriptase is inactivated
Animal derived additives	none

Storage and stability

Stable at -15 to -25°C until the control date printed on the label. Repeated freezing and thawing should be avoided.

Applications

- Synthesis of first strand cDNA for use in subsequent amplification reactions on conventional thermal cyclers and real-time instruments, *e.g.* LightCycler Instrument (RT-PCR)
- RT-PCR of GC-rich RNA templates with high secondary structure
- Cy3, Cy5, DIG, Biotin and aminoallyl labeling during cDNA synthesis
- Retrieve and clone the 5' and 3' termini of mRNA by RACE
- Generate cDNA libraries with large inserts
- Dideoxy DNA sequencing
- RNA sequencing
- 3'end labeling of DNA fragments
- Generation of ss probes for genomic footprints

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Roche

For life science research only. Not for use in diagnostic procedures.
FOR *IN VITRO* USE ONLY.

C. therm. Polymerase One-Step RT-PCR System

Cat. No. 2 016 338 50 reactions
Cat. No. 2 016 346 250 reactions

Store at −15 to −25° C

Instruction Manual

Version 1, June 1999

Reverse Transcription with *C. therm.* Polymerase One Step RT-PCR System has the following advantages:

- The high reaction temperature between 60° C and 70° C allows increased specificity of primer hybridization and subsequent extension compared to other enzymes commonly used for reverse transcription.
- *C. therm.* Polymerase One Step RT-PCR System minimizes problems encountered with secondary structures in RNA due to the improved performance in reverse transcription at higher temperatures and due to the use of DMSO in the RT-PCR buffer which further reduces the stability of RNA hairpin structures.
- The negative effect of manganese ions on the fidelity of DNA synthesis has been documented (4, 5). *C. therm.* Polymerase uses magnesium as a cofactor in cDNA synthesis and therefore has a fidelity rate more than two fold higher than that of Tth DNA polymerase.
- *C. therm.* Polymerase One Step RT-PCR System allows the amplification of fragments by RT-PCR up to 3 kb.
- Reverse transcription with *C. therm.* Polymerase and amplification with Taq DNA polymerase allows incorporation of dUTP (Cat. No.: 1 934 554) and decontamination with Uracil DNA Glycosylase (Cat. No.: 1 775 367).
- *C. therm.* Polymerase does not show any inhibition by the presence of high amounts of non specific RNA.